

Patent claims

1. Method for the identification of biomolecules in variant libraries of biomolecules comprising the steps:

5 a) Production of a variant library, consisting of a number of variants (B_0) of gene sequences coding for the biomolecule, and

b) Division of the variant library into a number of compartments (W_0), which is at least by a factor of ten smaller than the number of variants in the variant library (B_0),

10 Whereas each compartment contains a partial library which contains $K_0=B_0/W_0$ variants,

c) Production of biomolecules in the compartments and testing of the biomolecules obtained in the single compartments for a specified property (phenotype), whereas from the observed phenotype no direct conclusions on the genotype can be made,

15 d) Selection of at least one compartment, which contains biomolecules fulfilling the wanted properties,

e) Division of the partial library contained in the selected compartment into further compartments and

20 f) n-fold repetition of the steps c) to e) until in every compartment maximally only one variant ($K_n \leq 1$) of the gene sequence coding for the biomolecule is contained.

2. The method of claim 1, wherein the wanted property is a biocatalytic activity.

- 25 3. The method of claim 1 or 2, wherein in step c) also an amplification of the partial library takes place in the compartments up to an number of individuals $V_0(x)$ at the point in time x per compartment, whereas the number of individuals $V_0(x)$ divided by the number of clones per compartment K_0 gives the amplification factor $F_0(x)$ per clone.

4. The method of one of the claims 1 to 3, wherein in step e) the division is carried out under dilution of the partial library by means of factor $F_0(x)$, so that in a given volume every clone contained in the compartment is statistically present up to a number $X_0 < W_1$, this volume is divided up in a number of new compartments W_1 , whereas the new number of clones per compartment amounts to $K_1 = X_0 * K_0 / W_1$.

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5. The method of one of the claims 1 to 4, wherein the variant library contains 10^3 to 10^{15} variants of the gene sequence of the biomolecule.

6. The method of one of the claims 1 to 5, wherein in step b) the variant library is divided up in 10^1 to 10^4 compartments.

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7. The method of one of the claims 1 to 6, wherein in step b) the variant library is transferred into an organism before division.

8. The method of claim 7, wherein in step c) the culture of the organism after division is amplified to a number of organisms of 10^8 to 10^9 per compartment.

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9. The method of claim 7 or 8, wherein the organisms also conduct the production of the biomolecules.

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10. The method of one of the claims 7 to 9, wherein the partial libraries in the compartments are re-isolated from the organisms and the production of the biomolecules is conducted by cell-free systems.

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11. The method of one of the claims 1 to 6, wherein the amplification of the partial libraries and the production of the biomolecules is conducted by cell-free systems.

12. The method of one of the claims 1 to 11, wherein the variant library consists of DNA-plasmids, which contain the gene sequence coding for the biomolecule.

13. The method of one of the claims 1 to 11, wherein the variant library consists of linear nucleic acid molecules, which contain the gene sequence coding for the biomolecule.

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14. The method of one of the claims 1 to 13, wherein the biomolecules are enzymes or ribozymes or other biomolecules, which exhibit a biocatalytic activity.

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15. The method of one of the claims 1 to 14, wherein the test for a biocatalytic activity is conducted with physical detection methods, like preferentially the UV/VIS-spectroscopy, the fluorescence spectroscopy or the fluorescence-correlation-spectroscopy.